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**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

Applicant: Usha GOWSWAMI et al.  
Title: PROBES FOR MYCTOPHID FISH AND A  
METHOD FOR DEVELOPING THE SAME  
Prior Appl. No.: 09/782,604  
Prior Appl. Filing Date: February 14, 2001  
Examiner: Unassigned  
Art Unit: Unassigned

**PRELIMINARY AMENDMENT**

Commissioner for Patents  
Washington, D.C. 20231

Sir:

Prior to examination Applicants respectfully request that the above-identified application be amended as follows:

**In the Specification:**

Please amend the specification as follows:

On Page 13, delete the first paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

In still another embodiment the primer set (forward and backward primers) used for amplification and detection of Cyt b gene contains oligonucleotides with the sequences (SEQ ID NOS 1-2):

CYT 1:	5'	TGA YTT GAA RAA CCA YCG TTG	3'
CYT 2:	5'	CTC CAR TCT TCG RYT TAC AAG	3'

On Page 13, delete the second paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

In another embodiment (forward and backward primers) used for reamplification and detection of Cyt b gene contains oligonucleotides with the sequences (SEQ ID NOS 3 and 2):

CBI-L: 5' CCA TCC AAC ATC TCA GCA TGA TGA AA 3'  
CYT 2: 5' CTC CAR TCT TCG RYT TAC AAG 3'

On Page 13, delete the third paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

In another embodiment the primer set (forward and backward primers) used for PCR amplification and detection of D-Loop gene contains oligonucleotides with the sequences:

PRO-L : 5' CTA CC 3'  
D-LOOP H: 5' CCT GAA GTA GGA ACC AGA TG 3' (SEQ ID NO 4)

On Page 13, delete the fourth paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

In another embodiment (forward and backward primers) used for PCR amplification of ITS2 gene contains oligonucleotides with the sequences SEQ ID NOS 5-6):

ITS2 -F : 5' CTA CGC CTG TCT GAG TGT C 3'  
ITS2 -R : 5' ATA TGC TTA AAT TCA GCG GG 3'

On Page 13, delete the fifth paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

In yet another embodiment the primer set (forward and backward primers) used for PCR amplification of Rhodopsin gene Rod contains oligonucleotides with the sequences SEQ ID NOS 7-8):

ROD-R: 5' TCT TTC CGC AGC ACA ACG TGG 3'  
ROD-F: 5' CAT ATG AAT ACC CTC AGT ACT ACC 3'

On Page 13, delete the sixth paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

In still another embodiment the primer set (forward and backward primers) used for PCR amplification of 12S RNA gene contains oligonucleotides with the sequences (SEQ ID NOS 9-10):

12 SA-L: 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3'  
12 SB-H : 5 ' AGA GTG ACG GGC GGT GTG T 3'

On Page 14, delete the first paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

In another embodiment the primer set (forward and backward primers) used for PCR amplification of 16S RNA gene contains oligonucleotides with the sequences (SEQ ID NOS 11-12):

16 SAR -L: 5' CGC CTG TTT ATC AAA AAC AT 3'  
16 SBR-H : 5 ' CCG GTC TGA ACT CAG ATC ACG T 3'

On Page 14, delete the last paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The invention also relates to specific DNA sequences for the cloned DNA probe inserts for the Cyt b , D-Loop, Rod, ITS2 genes. The invention provides species specific primer sequences for amplification and detection of Cyt b , D-Loop, Rod, ITS2 , 12S RNA and 16 S RNA genes of *Stenobranchius leucopsarus* (SLMB) myctophid fish. The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of *Stenobranchius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 13-14):

12S-H 5' CCC ACT CAC TGC TAA CTC C 3'  
12S-L 5' GGC TAA CTA CAA TCA TCT GCT 3'

On Page 15, delete the first paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA -L of *Stenobrachius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 15-16):

16S-H	5'	TAC GCA TAA CGG CTC TGG	3'
16S-L	5'	CTA CTA CAC CTC AAC TAC ATC T	3'

On Page 15, delete the second paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer Cyt -H and Cyt -L of *Stenobrachius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 17-18):

Cyt-H	5'	GCT CGG GCT GCT GGA ATC TT	3'
Cyt-L	5'	CAA CCT CAT CTG TCG TAA AC	3'

On Page 15, delete the third paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer ITS2 -H and ITS2 -L (Forward ) of *Stenobrachius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 19-20):

ITS2-H	5'	ATA CTC TGC GGA CAT ACT TGA CTG	3'
ITS2-F	5'	ACT TGA CTG ACC TTC TTA CT	3'

On Page 15, delete the fourth paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer Pro-L and D Loop -H of *Stenobrachius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 21-22):

Pro-L	5'	CAG TCT CGT CAA ACC AAG TCA AAC	3'
D loop-H	5'	ATA ATC ATC CAG CAT AAA CAC AC	3'

On Page 15, delete the fifth paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer ROD -L and ROD-H of *Stenobranchius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 23-24):

ROD-L	5'	CCT GGT AGA GTT CGC CGT CA	3'
ROD-H5'		CGT GTT CCT TAT CAT TGT GCC T	3'

On Page 15, delete the sixth paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA-L of yet another myctophid *Lampanyctus regalis* ( LRMB) designed were such as (SEQ ID NOS 25-26):

16S-H	5'	TCG TAG TTC AGC AGT CAG	3'
16S-L	5'	CAC CAG CCA AGT ATG TTT CTC	3'

On Page 16, delete the first paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of yet another myctophid *Lampanyctus regalis* ( LRMB) designed were such as (SEQ ID NOS 27-28):

12S-H	5'	GCC TCC ATC ATC CCT CAC CTT AC	3'
12S-L	5'	CTA TTC GCC TCG CTC AGA C	3'

On Page 16, delete the second paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA -L of yet another myctophid *Diaphus theta* (DTMB) designed were such as SEQ ID NOS 29-30):

16S-H	5'	CTC CGT CCG TCT CGC CTC TG	3'
16S-L	5'	AAA TCC GCC CTT ATG TGT GTT C	3'

On Page 16, delete the third paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of yet another myctophid *Diaphus theta* (DTMB) designed were such as (SEQ ID NOS 31-32):

12S-H	5'	CAT CGG CTT GCT CTA TTC CTT G	3'
12S-L	5'	TCT ATC GGC GGC GTA TCA C	3'

On Page 16, delete the fourth paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA -L of yet another myctophid *Tarletonbaenia crenularis* (TCMB) designed were such as (SEQ ID NOS 33-34):

16S-H	5'	GGC GAT TCT ACG GCA CGG GCG	3'
16S-L	5'	AAA CTG GTC CTC AAC TAT GTC A	3'

On Page 16, delete the fifth paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of yet another myctophid *Tarletonbaenia crenularis* (TCMB) designed were such as (SEQ ID NOS 35-36):

12S-H	5'	CCG ATT CAG CCA CGA TTC CCT C	3'
12S-L	5'	CCT AAA GCC CAG ATA ACT ACA	3'

On Page 16, delete the sixth paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA -L of yet another myctophid *Protomyctophum crockeri* (PCMB) designed were such as (SEQ ID NOS 37-38):

16S-H	5'	CGT GTT CTG ATG ATG ATG TGC T	3'
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16S-L            5'        ATT CCT TCC TCT TAG TAT G            3'

On Page 17, delete the first paragraph; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of yet another myctophid *Protomyctophum crockeri* (PCMB) designed were such as (SEQ ID NOS 39-40):

12S-H            5'        GCT GAA CTT ACT ATG CCC TAC T            3'  
12S-L            5'        CCG ATT GAC GCC GAA CTA TG            3'

On Page 17, delete the paragraph entitled **Table 1**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 1 -** Forward primer (SEQ ID NO: 18) designed for cytochrome b gene of *Stenobrachius leucopsarus* ( slmb primer cyt L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 17, delete the paragraph entitled **Table 2**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 2 -** Backward primer (SEQ ID NO: 17) designed for cytochrome b gene of *Stenobrachius leucopsarus* (slmb primer cyt H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 17, delete the paragraph entitled **Table 3**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 3 -** Forward primer (SEQ ID NO: 20) designed for Internal Transcribed Spacer (ITS2) of *Stenobrachius leucopsarus* (slmb primer ITS2 F) with 5' to 3'

end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 17, delete the paragraph entitled **Table 4**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 4 -** Backward primer (SEQ ID NO: 19) designed for Internal Transcribed Spacer (ITS2) *Stenobrachius leucopsarus* (slmb primer ITS2-H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 17, delete the paragraph entitled **Table 5**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 5 -** Forward primer (SEQ ID NO: 21) designed for mitochondrial Control region d-Loop of *Stenobrachius leucopsarus* (slmb primer pro-L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 17, delete the paragraph entitled **Table 6**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 6 -** Backward primer (SEQ ID NO: 22) designed for mitochondrial Control region d-Loop *Stenobrachius leucopsarus* (slmb primer D loop -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 17, delete the paragraph entitled **Table 7**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 7 -** Forward primer (SEQ ID NO: 23) designed for Rhodopsin gene region of *Stenobrachius leucopsarus* (slmb primer ROD-L) with 5' to 3' end



sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 18, delete the paragraph entitled **Table 8**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 8 -** Backward primer (SEQ ID NO: 24) designed for Rhodopsin gene region of *Stenobranchius leucopsarus* (slmb primer ROD -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 18, delete the paragraph entitled **Table 9**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 9 -** Forward primer (SEQ ID NO: 26) designed for mitochondrial 16S ribosomal RNA region of *Lampanyctus regalis* (LRMB primer 16 S-L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 18, delete the paragraph entitled **Table 10**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 10 -** Backward primer (SEQ ID NO: 25) designed for mitochondrial 16S ribosomal RNA region of *Lampanyctus regalis* (LRMB primer 16 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 18, delete the paragraph entitled **Table 11**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 11 -** Forward primer (SEQ ID NO: 28) designed for mitochondrial 12 S ribosomal RNA region of *Lampanyctus regalis* (LRMB primer 12 S-L) with

5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 18, delete the paragraph entitled **Table 12**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 12** - Backward primer (SEQ ID NO: 27) designed for mitochondrial 12 S ribosomal RNA region of *Lampanyctus regalis* (LRMB primer 12 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 18, delete the paragraph entitled **Table 13**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 13** - Backward primer (SEQ ID NO: 29) designed for mitochondrial 16 S ribosomal RNA region of *Diaphus theta* (DTMB primer 16 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 18, delete the paragraph entitled **Table 14**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 14** - Forward primer (SEQ ID NO: 30) designed for mitochondrial 16 S ribosomal RNA region of *Diaphus theta* (DTMB primer 16 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 18, delete the paragraph entitled **Table 15**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 15** - Backward primer (SEQ ID NO: 31) designed for mitochondrial 12 S ribosomal RNA region of *Diaphus theta* (DTMB primer 12 S -H) with 5'

to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 18, delete the paragraph entitled **Table 16**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 16** - Forward primer (SEQ ID NO: 32) designed for mitochondrial 12 S ribosomal RNA region of *Diaphus theta* (DTMB primer 12 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 19, delete the paragraph entitled **Table 17**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 17** - Backward primer (SEQ ID NO: 33) designed for mitochondrial 16 S ribosomal RNA region of *Tarletonbeania crenularis* (TCMB primer 16 S - H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 19, delete the paragraph entitled **Table 18**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 18** - Forward primer (SEQ ID NO: 24) designed for mitochondrial 16 S ribosomal RNA region of *Tarletonbeania crenularis* (TCMB primer 16 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 19, delete the paragraph entitled **Table 19**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 19** - Backward primer (SEQ ID NO: 35) designed for mitochondrial 12 S ribosomal RNA region of *Tarletonbeania crenularis* (TCMB primer 12 S -

H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 19, delete the paragraph entitled **Table 20**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 20** - Forward primer (SEQ ID NO: 36) designed for mitochondrial 12 S ribosomal RNA region of *Tarletonbeania crenularis* (TCMB primer 12 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 19, delete the paragraph entitled **Table 21**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 21** - Backward primer (SEQ ID NO: 37) designed for mitochondrial 16 S ribosomal RNA region of *Protomyctophum crockeri* (PCMB primer 16 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 19, delete the paragraph entitled **Table 22**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 22** - Forward primer (SEQ ID NO: 38) designed for mitochondrial 16 S ribosomal RNA region of *Protomyctophum crockeri* (PCMB primer 16 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 19, delete the paragraph entitled **Table 23**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 23** - Backward primer (SEQ ID NO: 39) designed for mitochondrial 12 S ribosomal RNA region of *Protomyctophum crockeri* (PCMB primer 12 S

-H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Pages 19-20, delete the paragraph entitled **Table 24**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 24** - Forward primer (SEQ ID NO: 40) designed for mitochondrial 12 S ribosomal RNA region of *Protomyctophum crockeri* (PCMB primer 12 S - L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 20, delete the paragraph entitled **Table 25**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 25** - Backward primer (SEQ ID NO: 15) designed for mitochondrial 16 S ribosomal RNA region of *Stenobrachius leucopsarus* (SLMB primer 16 S - H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 20, delete the paragraph entitled **Table 26**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 26** - Forward primer (SEQ ID NO: 16) designed for mitochondrial 16 S ribosomal RNA region of *Stenobrachius leucopsarus* (SLMB primer 16 S - L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 20, delete the paragraph entitled **Table 27**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 27** - Backward primer (SEQ ID NO: 13) designed for mitochondrial 12 S ribosomal RNA region of *Stenobrachius leucopsarus* (SLMB primer 12 S

-H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

On Page 20, delete the paragraph entitled **Table 28**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Table 28** - Forward primer (SEQ ID NO: 14) designed for mitochondrial 12 S ribosomal RNA region of *Stenobrachius leucopsarus* (SLMB primer 12 S - L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

On Page 24, delete the paragraph entitled **Example 6**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Example 6:** PCR amplification using forward and backward D-Loop primers of *Stenobrachius leucopsarus*.

The PCR master mix (100 µl) comprised of Taq Buffer MgCl<sub>2</sub> free (10.0 µl), dNTP all the four nucleotides in the ratio of 1:1:1:1 (08.0 µl); D-Loop forward primer 01.0 µl with sequences ( PRO-L : 5' CTA CC 3' ), D-Loop backward 01.0 µl, with sequences (D-Loop H: 5' CCT GAA GTA GGA ACC AGA TG 3') (SEQ ID NO: 4); MgCl<sub>2</sub> ( 01.0 µl); Taq Polymerase (0.5 µl ); and ultrapure water ( 78.2 µl).

On Pages 24-25, delete the paragraph entitled **Example 7**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Example 7:**

As given in example 6, PCR amplification master mix was prepared using forward and backward 12 S RNA primers; 16 S RNA primers, Cyt b primers; ROD, ITS2 primers and DNA 0.3 µl of *Stenobrachius leucopsarus* was added individually to all tubes and amplified. The primers used were ROD-F: (SEQ ID NO: 8) 5' CAT ATG AAT ACC CTC AGT ACT ACC 3' and ROD-R: (SEQ ID NO: 7) 5' TCT TTC CGC AGC

ACA ACG TGG 3' for Rhodopsin DNA probe; 16SBR-H (SEQ ID NO: 12) 5' CCG GTC TGA ACT CAG ATC ACG T 3' and 16SAR-L (SEQ ID NO: 11) 5' CGC CTG TTT ATC AAA AAC AT 3' 16S for 16 S RNA gene probe; 12SA-L: (SEQ ID NO: 9) 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3' and 12SB-L: (SEQ ID NO: 10) 5' AGA GTG ACG GGC GGT GTG T 3' for 12S RNA gene probe and run for 35 cycles in DNA thermo cycler. (Each cycle consisted of 94 degree C for 45 Seconds, 48 degree for 45 seconds, and 72 degree C for 1 minute) and hold at 4 degree Centigrade.

On Page 25, delete the paragraph entitled **Example 8**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Example 8:**

Cytochrome b DNA probe was amplified by using Cyt 1: (SEQ ID NO: 1) 5' TGA YTT GAA RAA CCA YCG TTG 3' and Cyt 2: (SEQ ID NO: 2) 5' CTC CAR TCT TCG RYT TAC AAG 3' primers followed by reamplification by using CBI-L (SEQ ID NO: 3) 5' CCA TCC AAC ATC TCA GCA TGA TGA AA 3' and Cyt 2: (SEQ ID NO: 2) 5' CTC CAR TCT TCG RYT TAC AAG 3' primers. The DNA template was of *Stenobrachius leucopsarus* and run for 35 cycles in DNA thermo cycler. (Each cycle consisted of 94°C for 45 Seconds, 48 degree for 45 seconds, and 72 degree C for 1 minute) and hold at 4 degree Centigrade.

On Page 25, delete the paragraph entitled **Example 9**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Example 9:**

Similarly , primer ITS1F-ITS2R of Internal transcribed spacers was used for the nested PCR's ( ITS1-F: (SEQ ID NO: 41) 5' TTG TAC ACA CCG CCC GTC GC 3' and ITS2-R: (SEQ ID NO: 6) 5' ATA TGC TTA AAT TCA GCG GG 3') and amplified by PCR. Later the ITS2 was reamplified using primers ITS2-F: (SEQ ID NO: 5) 5' CTA CGC CTG TCT GAG TGT C 3' and

ITS2-R: (SEQ ID NO: 6) 5' ATA TGC TTA AAT TCA GCG GG 3'. The DNA template was of *Stenobranchius leucopsarus* myctophid fish and run for 35 cycles in DNA thermo cycler. (Each cycle consisted of 94°C for 45 Seconds, 48°C for 45 seconds, and 72°C for 1 minute) and hold at 4 degree Centigrade.

On Page 29, delete the paragraph entitled **Example 20**; and replace this paragraph with the following in accordance with 37 CFR §1.121. A marked up version showing changes is attached:

**Example 20 :**

PCR for confirmation that transformed bacteria has the plasmids with the D-Loop gene inserts.:

PCR amplification using forward and backward D-Loop primers of *Stenobranchius leucopsarus*.

The PCR master mix (100 µl ) comprised of Taq Buffer MgCl<sub>2</sub> free (10.0 µl ), dNTP all the four nucleotides in the ratio of 1:1:1:1 (08.0 µl); D-Loop forward primer 01.0 µl with sequences ( PRO-L : 5' CTA CC 3'), D-Loop backward 01.0 µl, with sequences (D-Loop H: 5' CCT GAA GTA GGA ACC AGA TG 3') (SEQ ID NO: 4); MgCl<sub>2</sub> (01.0 µl); Taq Polymerase (0.5 µl ); and ultrapure water ( 78.2 µl).

**In the Claims:**

In accordance with 37 C.F.R. § 1.121, please substitute for original claims 9-19, 29-31, 33-41 and 74-107 the following rewritten version of the same claims, as amended. The changes are shown explicitly in the attached "Version With Markings to Show Changes Made."

9. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for amplification and detection of Cyt b gene contains oligonucleotides with the sequences (SEQ ID NOS 1-2):

CYT 1:	5'	TGA YTT GAA RAA CCA YCG TTG	3'
CYT 2:	5'	CTC CAR TCT TCG RYT TAC AAG	3'



10. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for reamplification and detection of Cyt b gene contains oligonucleotides with the sequences (SEQ ID NOS 3 and 2):

CBI-L: 5' CCA TCC AAC ATC TCA GCA TGA TGA AA 3'  
CYT 2: 5' CTC CAR TCT TCG RYT TAC AAG 3'

11. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification and detection of D-Loop gene contains oligonucleotides with the sequences :

PRO-L : 5' CTA CC 3'  
D-LOOP H: 5' CCT GAA GTA GGA ACC AGA TG 3' (SEQ ID NO: 4)

12. (Amended) A method as claimed in claim 1 wherein the forward and backward primers used for PCR amplification of ITS2 gene were

ITS1 F : 5' TTG TAC ACA CCGCCCGTC GC 3' (SEQ ID NO: 41)  
ITS2 R : 5' ATA TGC TTA AAT TCA GCG GG 3' (SEQ ID NO: 6)

13. (Amended) A method as claimed in claim 1 wherein the forward and backward primers used for PCR reamplification of ITS2 gene from ITS1 F and ITS2 R PCR amplification were

ITS2 F: 5' CTA CGC CTG TCT GAG TGT C 3' (SEQ ID NO: 5)  
ITS2 R: 5' ATA TGC TTA AAT TCA GCG GG 3' (SEQ ID NO: 6)

14. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of Rhodopsin gene Rod contains oligonucleotides with the sequences (SEQ ID NOS 8 and 7):

ROD-F: 5' CAT ATG AAT ACC CTC AGT ACT ACC 3'  
ROD-R: 5' TCT TTC CGC AGC ACA ACG TGG 3'

15. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of 12S RNA gene contains oligonucleotides with the sequences (SEQ ID NOS 9-10):

12 SA-L: 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3'  
 12 SB-H : 5 ' AGA GTG ACG GGC GGT GTG T 3'

16. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of 16S RNA gene contains oligonucleotides with the sequences:

16 SAR -L: 5' CGC CTG TTT ATC AAA AAC AT 3' (SEQ ID NO: 11)  
 16 SBR-H : 5 ' CCG GTC TGA ACT CAG ATC ACG T 3' (SEQ ID NO: 12)

17. (Amended) A method as claimed in claim 1 wherein the forward and backward primers used for PCR amplification of Rhodopsin gene Rod were (SEQ ID NOS 8 and 7:

ROD-F: 5' CAT ATG AAT ACC CTC AGT ACT ACC 3'  
 ROD-R: 5' TCT TTC CGC AGC ACA ACG TGG 3'

18. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of 12S RNA gene were (SEQ ID NOS 9-10):

12 SA-L: 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3'  
 12 SB-H: 5 ' AGA GTG ACG GGC GGT GTG T 3'

19. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of 16S RNA gene were (SEQ ID NOS 11-12):

16 SAR -L: 5' CGC CTG TTT ATC AAA AAC AT 3'  
 SBR-H: 5 ' CCG GTC TGA ACT CAG ATC ACG T 3'

29. (Amended) A method claimed in claim 1 wherein the cycle sequencing primer for CYT b gene consisted of oligonucleotides with the sequence:

CYT 1: 5' TGA YTT GAA RAA CCA YCG TTG 3' (SEQ ID NO: 1)

30. (Amended) A method claimed in claim 1 wherein the cycle sequencing primer for CYT b gene consisted of oligonucleotides with the sequence:

CYT 2: 5' CTC CAR TCT TCG RYT TAC AAG 3' (SEQ ID NO: 2)

31. (Amended) A method claimed in claim 1 wherein the cycle sequencing primer for CYT b gene consisted of oligonucleotides with the sequence:

CBI-L: 5' CCA TCC AAC ATC TCA GCA TGA TGA AA 3' (SEQ ID NO: 3)

33. (Amended) A method claimed in claim 1 wherein the backward cycle sequencing primer for D-Loop region consisted of oligonucleotides with the sequence:

D-LOOP H: 5' CCT GAA GTA GGA ACC AGA TG 3' (SEQ ID NO: 4)

34. (Amended) A method as claimed in claim 1 wherein the forward primer used for cycle sequencing of ITS2 gene consisted of oligonucleotides with the sequence:

ITS 1 -F : 5' TTG TAC ACA CCG CCC GTC GC 3' (SEQ ID NO: 41)

35. (Amended) A method as claimed in claim 1 wherein the backward primer used for cycle sequencing of ITS2 gene consisted of oligonucleotides with the sequence:

ITS2 -R : 5' ATA TGC TTA AAT TCA GCG GG 3' (SEQ ID NO: 6)

36. (Amended) A method as claimed in claim 1 wherein the forward primer used for cycle sequencing of Rhodopsin gene Rod consisted of oligonucleotides with the sequence:

ROD-F: 5' CAT ATG AAT ACC CTC AGT ACT ACC 3' (SEQ ID NO: 8)

37. (Amended) A method as claimed in claim 1 wherein the backward primer used for cycle sequencing consisted of oligonucleotides with the sequence:

ROD-R: 5' TCT TTC CGC AGC ACA ACG TGG 3' (SEQ ID NO: 7)

38. (Amended) A method as claimed in claim 1 wherein the forward primer used for cycle sequencing of 12S RNA gene consisted of oligonucleotides with the sequence:

12 SA-L: 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3' (SEQ ID NO: 9)

39. (Amended) A method as claimed in claim 1 wherein the backward primer used for cycle sequencing of 12S RNA gene consisted of oligonucleotides with the sequence:

12 SB-H: 5' AGA GTG ACG GGC GGT GTG T 3' (SEQ ID NO: 10)

40. (Amended) A method as claimed in claim 1 wherein the forward primer used for cycle sequencing of 16S RNA gene consisted of oligonucleotides with the sequence:

16 SAR -L: 5' CGC CTG TTT ATC AAA AAC AT 3' (SEQ ID NO: 11)

41. (Amended) A method as claimed in claim 1 wherein the backward primer used for cycle sequencing of 16S RNA gene consisted of oligonucleotides with the sequence:

16 SBR-H: 5' CCG GTC TGA ACT CAG ATC ACG T 3' (SEQ ID NO: 12)

74. (Amended) The nucleotide base sequences of PSL CYTL (747 bp) comprising (SEQ ID NO: 42):

5'

CTTNCCCATT	TTGGGCGCTT	NGGCNCGCTN	CTCCNCGAGA	CTCTGCGTAN
TAATCCAANT	CNCTNCGGGC	CNCTCCCTAC	CANTNCNCTA	CACCNCAAAT
TNCAACCCNG	TTTCCTCATC	ANTCAACCAC	ATCTGTCGAA	AACNTCAACT
ACGGCTGACT	AATCCGAAAA	CATGCACGCT	AACGGTGCCT	CTTTCTTCTT
CATCTGTATT	TATCTNCNCN	TTGGANGAGG	ACTATNCTAC	GGATCCTACC

TCTACGAAGA	GACGTGAGGT	GTTGGTGTTA	TTCTTCTCCT	TCTAATAATG
ATGACTGCNT	TTGTTGGCTA	TGTGCTNCCC	NGAGGACAAA	TGTCCTTTTG
AGGTGCTACT	GTCATTACAA	NCCTACTCTC	TGCTGTNCCG	TNTGTTNGCG
GCNCTCTANT	TCAATGAATT	TGAGGTGGCT	TCTCCGTAAA	CACGCAACGC
TCACTCGTTT	CTTCGCNTTC	CACTTCTTGT	TCCCATTGTG	TGTCGCNGCT
ATAACCNNGG	TTCACCNGAT	TTNCCGACAT	CAAACAGGCT	CTAAANCCCC
CCCGGNTTGA	CTCCATACAA	CAAAACCCTC	CACCCTATTC	NCTATAAAAC
TCTAGGTTCTG	TGCCCGTATT	GGCTTACTTC	ATGNCTATTT	CCCNGNCGGA
GGGACNAAAA	TTCCTGCACC	CCCTCCCCNC	AAAATAAANA	ATGTGTCTNT
CCTACCANAA	AACAACNNAN	ACGGGGTNTG	CNCTTCCATC	ATCCACN 3'

75. (Amended) The nucleotide base sequences of PSLITS2F comprises:  
(225BP) (SEQ ID NO: 43)

5'

TCTACGATCT	ACCGGCNTTT	NNTGTGGAAA	GACGATCATG
CATTTATGTG	TGTCTTTCTA	TGGATTTGAA	CCGTGTGGTA
CGTCTTTGCG	TACTGCTTGG	AAGGCTCAAC	TTGCTTCTGT
CCTTCTCTTG	CAGTCTCGCA	CTGTCTATGC	AACGTGTTCT
ACTTCGACTT	CTGTCGAAAA	ATCTTACTTT	TGACCTCAGA
TCAGACAAGA	CTACCCGCTG	AATT	3'

76. (Amended) The nucleotide base sequences of PSL PROL comprises:  
(750 BP) (SEQ ID NO: 44)

5'

CCTTTTCGGN	ATAGGCCCAN	CTCAAATGAA	TTCCTTCTCT
CCTGGTCCAA	GCCCAAAGTG	TGGACGGCAG	GTTGACAATG
GTTACAAATC	GTGACAAATC	GGCTACATAA	TTGCCGATAG
CGATGTCGTC	AAACCAAGTC	AAACAATGGC	CGATGTATAT
CGGCCAAACC	CATATATGGG	TCTGGCTGTA	GTTTGTGTTG
AGCAACGTCA	CACCAAGTGC	TGGTCAGCAT	ATAAGATGTT
GACATCTTGC	AACATCTTAC	CCACAGACAG	ACAGTTACGG
CTGCTTACGA	ANGGCGCTAG	TGTTGTGGTG	AGAAACGAAG
ATACATACGT	CAAACAGACG	CCGGTGCACT	TGAAGACACT

GTTTGAAGGT	GCCGCACTAC	TTGACAGACA	GCCCATGATG
CGCTGGACAG	TGACCAAAGC	TACNGGAGGA	CCANATGGAA
ATCCTGTTGG	CGTTGCCGTG	GGACTCAAGT	TGTACACTTT
TGGATGGTTG	ATCACTANAN	CCGCTGCCGG	GAGAAGCACT
CGCTCCTGGT	TCACTAATCA	GATTGAGGTT	AACCANATTG
ANGTAAACAT	CTTCAACACA	GTGTCTTTAT	GCTGGATGAA
ATTNAGCCCA	CNGGACACCA	NAAAAGAATT	NCCNCTGGTT
CTNNCGGGGG	NCCCCNNNAA	CGNNTNTTCC	CCTTNTCTCN
NNNGCGGNGA	AGTTNCCCCC	CCCCACTNAN	NTCTTCCTTC
AANANNTTTC	CNCCNNNAGA	GGTTTTCCCN	3'

77. (Amended) The nucleotide base sequences of ROD PSL SLMB comprises: (748 BP) (SEQ ID NO: 45)

5'			
CCTGGTAGGG	TTCCCCGTCA	ACTTCCTCAC	ACTGTACCTC
ACNTTCGAGC	ACAAGAAGCT	ACTAACCCCC	TTAAACTACA
TCCTGCTCAA	CCTGGCGGTC	GGAGACCTCC	TGATGGTGTA
AGGAGGGTTC	ACCACCACCA	TCTACACCTC	CATGCACGGC
TACTTCGTCC	TAGGGAAACT	GGGCTGCGCC	ATCGAAGGTT
TCATGGCCAC	CCATGGTGGT	CAGGTCGCCC	TTTGGTCCCT
GGTGGTTTTG	GCCGTGGAAA	GGTGGCTGGT	CGTCTGCAAN
CCCATCTCCA	GCTTCCGCTT	CCAGGAGTCC	CACTCCCTCA
TGGGCCTGGC	CGTGACCTGG	GTGATGGCGA	CGGCTTGTTT
TGTGCCCCCC	CTGGGTCGGC	TGGTCTCGCT	ACATCCCAGA
AGGCATGCAG	TGCTCATGCG	GAATGGACTA	CTACACTCCC
GCGCCGGGCG	TCAACAATGA	ATCCTACGTN	GTGTACATGT
TCNTCANAAA	AANAATNGGA	CCNCNGGGCG	ATCATNTTGN
TANGNNAAGG	CCAGNTGNTG	NGAGCAGTCA	AGGCGGCCGC
CGCCGCCAG	CAAGAGTCCG	AGACCACCCA	GAGGGCCGAG
AGGGAAGTCA	CCCGNATGGT	NATNANGATG	GTNATNTCNT
TCNTGGTAAG	NAGGGNGCCA	NACGCCAGCG	TGGCCTGGTG
GATCTTNGN	AACCAGGGNG	CAGAATTAGG	CCCNGTNTTC

ATGACCCTGC CGGCNTTCTT TGCCAAGA 3'

78. (Amended) A method as claimed in claim 1 wherein FORWARD ( L) primers of CYT b gene region for myctophid fish *Stenobranchius leucopsarus* is an oligonucleotide comprising (SEQ ID NO: 18):

5' CAA CCT CAT CTG TCG TAA AC 3'

and having the following characteristics:

- i. is a 20-mer DNA oligonucleotide ( sense),
- ii. has melting temperature of 56.4 degree celcius,
- iii. has a molecular weight of 6101.0,
- iv. has no hairpin loops,
- v. has no single dimers,
- vi. has no other dimers,
- vii. has no single bulge loops or internal loops, and
- viii. has no palindromes.

79. (Amended) A method as claimed in claim 1 wherein BACKWARD (H) primer of CYT b gene region for myctophid fish *Stenobranchius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 17):

5' GCT CGG GCT GCT GGA ATC TT 3'

and having the following characteristics:

- i. is a 20-mer DNA
- ii. is an antisense oligonucleotide
- iii. has a melting point of 70.8 degree celcius.
- iv. has a molecular weight of 6220.1.
- v. has no hairpin loops, no single bulge loops, no other internal loops, no single internal loops, no other bulge loops or palindromes.
- vi. no single dimers or other dimers.

80. (Amended) A method as claimed in claim 1 wherein forward primer of ITS2 F gene region for myctophid fish *Stenobranchius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 20):

5' ACT TGA CTG ACC TTC TTA CT 3'

and having the following characteristics:

- i. is a 20-mer sense oligonucleotide,
- ii. has a melting point of 51.3 degree celcius,
- iii. has a molecular weight of 6098.0,
- iv. has no palindromes, loops and dimers,

81. (Amended) A method as claimed in claim 1 wherein forward primer of ITS2 H gene region for myctophid fish *Stenobranchius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 19):

5' ATA CTC TGC GGA CAT ACT TGA CTG 3'

and having the following characteristics:

- i. is a 24-mer antisense oligonucleotide,
- ii. has a melting point of 65.4 degree celcius.
- iii. has a molecular weight of 7407.9.
- iv. has no palindromes, loops and dimers.

82. (Amended) A method as claimed in claim 1 wherein forward primer of pro-L for myctophid fish *Stenobranchius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 21):

5' CAG TCT CGT CAA ACC AAG TCA AAC 3'

and having the following characteristics:

- i. is a 24-mer sense oligonucleotide
- ii. has a melting point of 67.8 degree celcius.
- iii. has a molecular weight of 7354.9.
- iv. has no palindromes, loops and dimers.

83. (Amended) A method as claimed in claim 1 wherein backward primer for Dloop for mitochondrial control region (dloop H) gene region for myctophid fish *Stenobranchius leucopsarus* is an oligonucleotide comprising (SEQ ID NO: 22):

5' ATA ATC ATC CAG CAT AAA CAC AC 3'

and having the following characteristics:

- i. is a 23-mer antisense oligonucleotide,
- ii. has a melting point of 61.2 degree celcius.



- iii. has a molecular weight of 7033.7.
- iv. has no palindromes, loops and dimers.

84. (Amended) A method as claimed in claim 1 wherein the FORWARD primer ( ROD- L) for Rhodopsin gene region of myctophid fish *Stenobrachius leucopsarus* is an oligonucleotide comprising (SEQ ID NO: 23):

5' CCT GGT AGA GTT CGC CGT CA 3'

and having the following characteristics:

- i. is a 20-mer sense oligonucleotide
- ii. has a melting point of 67.4 degree celcius.
- iii. has a molecular weight of 6189.0.
- iv. has no palindromes, loops and dimers.

85. (Amended) A method as claimed in claim 1 wherein the backward primer ( ROD- H) for Rhodopsin gene region of myctophid fish *Stenobrachius leucopsarus* is an oligonucleotide comprising (SEQ ID NO: 24):

5' CGT GTT CCT TAT CAT TGT GCC T 3'

and having the following characteristics:

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 66.4 degree celcius.
- iii. has a molecular weight of 6738.4.
- iv. has no palindromes, loops and dimers.

86. (Amended) A method as claimed in claim 1 wherein the forward primer of 16S-L of the myctophid fish *Lampanyctus regalis* is an oligonucleotide comprising (SEQ ID NO: 26):

5' CAC CAG CCA AGT ATG TTT CTC 3'

and having the following characteristics:

- i. is a 21-mer sense oligonucleotide
- ii. has a melting point of 61.5 degree celcius.
- iii. has a molecular weight of 6421.4.
- iv. has no palindromes, loops and dimers.

87. (Amended) A method as claimed in claim 1 wherein the backward primer of 16s rRNA of myctophid fish *Lampanyctus regalis* is an oligonucleotide comprising (SEQ ID NO: 25):

5' TCG TAG TTC AGC AGT CAG 3'

and having the following characteristics:

- i. is a 18-mer antisense oligonucleotide
- ii. has a melting point of 51.2 degree celcius.
- iii. has a molecular weight of 5594.7.
- iv. has no palindromes, hairpin loops and dimers.

88. (Amended) A method as claimed in claim 1 wherein the forward primer 16S-L of myctophid fish *Lampanyctus regalis* is an oligonucleotide comprising (SEQ ID NO: 28):

5' CTA TTC GCC TCG CTC AGA C 3'

and having the following characteristics:

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 62.1 degree celcius.
- iii. has a molecular weight of 5779.8.
- iv. has no palindromes, hairpin loops and dimers.

89. (Amended) A method as claimed in claim 1 wherein a primer 12S-H for *Lampanyctus regalis* (LRMB) is an oligonucleotide comprising (SEQ ID NO: 27):

5' GCC TCC ATC ATC CCT CAC CTT AC 3'

and having the following characteristics:

- i. is a 23-mer antisense oligonucleotide
- ii. has a melting point of 70.8 degree celcius.
- iii. has a molecular weight of 6895.5
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

90. (Amended) A method as claimed in claim 1 wherein the primer 12S-L for *Lampanyctus regalis* (LRMB) is an oligonucleotide comprising (SEQ ID NO: 28):

5' CTA TTC GCC TCG CTC AGA C 3'

and having the following characteristics:

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 62.1 degree celcius.
- iii. has a molecular weight of 5779.8
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

91. (Amended) A method as claimed in claim 1 wherein 16S-L forward primer for *Diaphus theta* (DTMB) is an oligonucleotide comprising (SEQ ID NO: 30):

5' AAA TCC GCC CTT ATG TGT GTT C 3'

and having the following characteristics:

- i. is a 22-mer sense oligonucleotide
- ii. has a melting point of 67.9 degree celcius.
- iii. has a molecular weight of 6756.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

92. (Amended) A method as claimed in claim 1 wherein 16S-H backward primer for *Diaphus theta* (DTMB) is an oligonucleotide comprising (SEQ ID NO: 29):

5' CTC CGT CCG TCT CGC CTC TG 3'

and having the following characteristics:

- i. is a 20-mer antisense oligonucleotide
- ii. has a melting point of 71.7 degree celcius.
- iii. has a molecular weight of 6052.0
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

93. (Amended) A method as claimed in claim 1 wherein 12S-H forward primer for *Diaphus theta* (DTMB) is an oligonucleotide comprising (SEQ ID NO: 31):

5' CAT CGG CTT GCT CTA TTC CTT G 3'

and having the following characteristics:

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 68.8 degree celcius.

- iii. has a molecular weight of 6723.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

94. (Amended) A method as claimed in claim 1 wherein 12S-L forward primer for *Diaphus theta* (DTMB) is an oligonucleotide comprising (SEQ ID NO: 32):

5' TCT ATC GGC GGC GTA TCA C 3'

and having the following characteristics:

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 65.8 degree celcius.
- iii. has a molecular weight of 5859.8
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

95. (Amended) A method as claimed in claim 1 wherein 16S-H primer for *Tarletonbeania crenularis* (TCMB) is an oligonucleotide comprising (SEQ ID NO: 33):

5' GGC GAT TCT ACG GCA CGG GCG 3'

and having the following characteristics:

- i. is a 21-mer antisense oligonucleotide
- ii. has a melting point of 80.4 degree celcius.
- iii. has a molecular weight of 6568.3
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

96. (Amended) A method as claimed in claim 1 wherein 16S-L forward primer for *Tarletonbeania crenularis* (TCMB) is an oligonucleotide comprising (SEQ ID NO: 34):

5' AAA CTG GTC CTC AAC TAT GTC A 3'

and having the following characteristics:

- i. is a 22-mer sense oligonucleotide
- ii. has a melting point of 60.7 degree celcius.
- iii. has a molecular weight of 6758.5

iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

97. (Amended) A method as claimed in claim 1 wherein 16S-H backward primer for *Tarletonbeania crenularis* ( TCMB) is an oligonucleotide comprising (SEQ ID NO: 33):

5' GGC GAT TCT ACG GCA CGG GCG 3'

and having the following characteristics:

- i. is a 21-mer antisense oligonucleotide
- ii. has a melting point of 80.4 degree celcius.
- iii. has a molecular weight of 6568.3
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

98. (Amended) A method as claimed in claim 1 wherein 12S-H backward primer for *Tarletonbeania crenularis* ( TCMB) is an oligonucleotide comprising (SEQ ID NO: 35):

5' CCG ATT CAG CCA CGA TTC CCT C 3'

and having the following characteristics:

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 74.6 degree celcius.
- iii. has a molecular weight of 6671.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

99. (Amended) A method as claimed in claim 1 wherein 12S-L forward primer for *Tarletonbeania crenularis* ( TCMB) is an oligonucleotide comprising (SEQ ID NO: 42):

5' CCT AAA GCC CAG ATA ACT ACA 3'

- i. is a 21-mer sense oligonucleotide
- ii. has a melting point of 59.2 degree celcius.
- iii. has a molecular weight of 6432.3

iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

100. (Amended) A method as claimed in claim 1 wherein 16S-H backward primer for *Protomyctophum crockeri* (PCMB) is an oligonucleotide comprising (SEQ ID NO: 37):

5' CGT GTT CTG ATG ATG ATG TGC T 3'

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 64.7 degree celcius.
- iii. has a molecular weight of 6867.5
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

101. (Amended) A method as claimed in claim 1 wherein 16S-L forward primer for *Protomyctophum crockeri* (PCMB) is an oligonucleotide comprising (SEQ ID NO: 38):

5' ATT CCT TCC TCT TAG TAT G 3'

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 49.5 degree celcius.
- iii. has a molecular weight of 5799.8
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

102. (Amended) A method as claimed in claim 1 wherein 12S-H backward primer for *Protomyctophum crockeri* (PCMB) is an oligonucleotide comprising (SEQ ID NO: 39):

5' GCT GAA CTT ACT ATG CCC TAC T 3'

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 60.3 degree celcius.
- iii. has a molecular weight of 6725.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

103. (Amended) A method as claimed in claim 1 wherein 12S-L forward primer for *Protomyctophum crockeri* (PCMB) is an oligonucleotide comprising (SEQ ID NO: 40):

5' CCG ATT GAC GCC GAA CTA TG 3'

- i. is a 20-mer sense oligonucleotide
- ii. has a melting point of 68.1 degree celcius.
- iii. has a molecular weight of 6182.1
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

104. (Amended) A method as claimed in claim 1 wherein 16S-H backward primer for *Stenobrachius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 15):

5' TAC GCA TAA CGG CTC TGG 3'

- i. is a 18-mer DNA oligonucleotide ( Antisense)
- ii. has a melting point of 61.4 degree celcius.
- iii. has a molecular weight of 5579.7
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

105. (Amended) A method as claimed in claim 1 wherein 16S-L forward primer for *Stenobrachius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 16):

5' CTA CTA CAC CTC AAC TAC ATC T 3'

- i. is a 22-mer sense oligonucleotide
- ii. has a melting point of 52.4 degree celcius.
- iii. has a molecular weight of 6638.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

106. (Amended) A method as claimed in claim 1 wherein 12S-H forward primer for *Stenobrachius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 13):

5' CCC ACT CAC TGC TAA CTC C 3'

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 58.4 degree celcius.
- iii. has a molecular weight of 5708.8
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

107. (Amended) A method as claimed in claim 1 wherein 12S-L forward primer for *Stenobrachius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 14:

5' GGC TAA CTA CAA TCA TCT GCT 3'

- i. is a 21-mer sense oligonucleotide
- ii. has a melting point of 58.5 degree celcius.
- iii. has a molecular weight of 6445.2
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.



**REMARKS**

Applicants respectfully request that the foregoing amendments be made prior to examination of the present application.

After amending the claims as set forth above, claims 107 are now pending in this application.

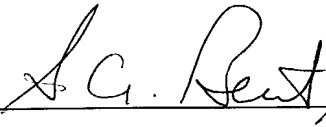
Applicants believe that the present application is now in condition for allowance. Favorable consideration of the application as amended is respectfully requested.

The Examiner is invited to contact the undersigned by telephone if it is felt that a telephone interview would advance the prosecution of the present application.

Respectfully submitted,

Date 13 September 2001

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**Version With Markings to Show Changes Made**

**Marked up replacement paragraphs:**

Page 13, first paragraph:

In still another embodiment the primer set (forward and backward primers) used for amplification and detection of Cyt b gene contains oligonucleotides with the sequences (SEQ ID NOS 1-2):

CYT 1:	5'	TGA YTT GAA RAA CCA YCG TTG	3'
CYT 2:	5'	CTC CAR TCT TCG RYT TAC AAG	3'

Page 13, second paragraph:

In another embodiment (forward and backward primers) used for reamplification and detection of Cyt b gene contains oligonucleotides with the sequences (SEQ ID NOS 3 and 2):

CBI-L:	5'	CCA TCC AAC ATC TCA GCA TGA TGA AA	3'
CYT 2:	5'	CTC CAR TCT TCG RYT TAC AAG	3'

Page 13, third paragraph:

In another embodiment the primer set (forward and backward primers) used for PCR amplification and detection of D-Loop gene contains oligonucleotides with the sequences:

PRO-L :	5'	CTA CC	3'
D-LOOP H:	5'	CCT GAA GTA GGA ACC AGA TG	3' <u>(SEQ ID NO 4)</u>

Page 13, fourth paragraph:

In another embodiment (forward and backward primers) used for PCR amplification of ITS2 gene contains oligonucleotides with the sequences SEQ ID NOS 5-6):

ITS2 -F :	5'	CTA CGC CTG TCT GAG TGT C	3'
ITS2 -R :	5'	ATA TGC TTA AAT TCA GCG GG	3'

Page 13, fifth paragraph:

In yet another embodiment the primer set (forward and backward primers) used for PCR amplification of Rhodopsin gene Rod contains oligonucleotides with the sequences SEQ ID NOS 7-8):

ROD-R: 5' TCT TTC CGC AGC ACA ACG TGG 3'  
ROD-F: 5' CAT ATG AAT ACC CTC AGT ACT ACC 3'

Page 13, sixth paragraph:

In still another embodiment the primer set (forward and backward primers) used for PCR amplification of 12S RNA gene contains oligonucleotides with the sequences (SEQ ID NOS 9-10):

12 SA-L: 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3'  
12 SB-H : 5' AGA GTG ACG GGC GGT GTG T 3'

Page 14, first paragraph:

In another embodiment the primer set (forward and backward primers) used for PCR amplification of 16S RNA gene contains oligonucleotides with the sequences (SEQ ID NOS 11-12):

16 SAR -L: 5' CGC CTG TTT ATC AAA AAC AT 3'  
16 SBR-H : 5' CCG GTC TGA ACT CAG ATC ACG T 3'

Page 14, last paragraph:

The invention also relates to specific DNA sequences for the cloned DNA probe inserts for the Cyt b , D-Loop, Rod, ITS2 genes. The invention provides species specific primer sequences for amplification and detection of Cyt b , D-Loop, Rod, ITS2 , 12S RNA and 16 S RNA genes of *Stenobranchius leucopsarus* (SLMB) myctophid fish. The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of *Stenobranchius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 13-14):

12S-H 5' CCC ACT CAC TGC TAA CTC C 3'  
12S-L 5' GGC TAA CTA CAA TCA TCT GCT 3'

Page 15, first paragraph:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA -L of *Stenobrachius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 15-16):

16S-H	5'	TAC GCA TAA CGG CTC TGG	3'
16S-L	5'	CTA CTA CAC CTC AAC TAC ATC T	3'

Page 15, second paragraph:

The sequences of the species specific primer Cyt -H and Cyt -L of *Stenobrachius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 17-18):

Cyt-H	5'	GCT CGG GCT GCT GGA ATC TT	3'
Cyt-L	5'	CAA CCT CAT CTG TCG TAA AC	3'

Page 15, third paragraph:

The sequences of the species specific primer ITS2 -H and ITS2 -L (Forward ) of *Stenobrachius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 19-20):

ITS2-H	5'	ATA CTC TGC GGA CAT ACT TGA CTG	3'
ITS2-F	5'	ACT TGA CTG ACC TTC TTA CT	3'

Page 15, fourth paragraph:

The sequences of the species specific primer Pro-L and D Loop -H of *Stenobrachius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 21-22):

Pro-L	5'	CAG TCT CGT CAA ACC AAG TCA AAC	3'
D loop-H	5'	ATA ATC ATC CAG CAT AAA CAC AC	3'

Page 15, fifth paragraph:

The sequences of the species specific primer ROD -L and ROD-H of *Stenobrachius leucopsarus* (SLMB) designed were such as (SEQ ID NOS 23-24):

ROD-L	5'	CCT GGT AGA GTT CGC CGT CA	3'
ROD-H5'		CGT GTT CCT TAT CAT TGT GCC T	3'

Page 15, sixth paragraph:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA-L of yet another myctophid *Lampanyctus regalis* ( LRMB) designed were such as (SEQ ID NOS 25-26):

16S-H	5'	TCG TAG TTC AGC AGT CAG	3'
16S-L	5'	CAC CAG CCA AGT ATG TTT CTC	3'

Page 16, first paragraph:

The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of yet another myctophid *Lampanyctus regalis* ( LRMB) designed were such as (SEQ ID NOS 27-28):

12S-H	5'	GCC TCC ATC ATC CCT CAC CTT AC	3'
12S-L	5'	CTA TTC GCC TCG CTC AGA C	3'

Page 16, second paragraph:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA -L of yet another myctophid *Diaphus theta* (DTMB) designed were such as SEQ ID NOS 29-30):

16S-H	5'	CTC CGT CCG TCT CGC CTC TG	3'
16S-L	5'	AAA TCC GCC CTT ATG TGT GTT C	3'

Page 16, third paragraph:

The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of yet another myctophid *Diaphus theta* (DTMB) designed were such as (SEQ ID NOS 31-32):

12S-H	5'	CAT CGG CTT GCT CTA TTC CTT G	3'
12S-L	5'	TCT ATC GGC GGC GTA TCA C	3'

Page 16, fourth paragraph:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA -L of yet another myctophid *Tarletonbaenia crenularis* (TCMB) designed were such as (SEQ ID NOS 33-34):

16S-H	5'	GGC GAT TCT ACG GCA CGG GCG	3'
16S-L	5'	AAA CTG GTC CTC AAC TAT GTC A	3'

Page 16, fifth paragraph:

The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of yet another myctophid *Tarletonbaenia crenularis* (TCMB) designed were such as (SEQ ID NOS 35-36):

12S-H	5'	CCG ATT CAG CCA CGA TTC CCT C	3'
12S-L	5'	CCT AAA GCC CAG ATA ACT ACA	3'

Page 16, sixth paragraph:

The sequences of the species specific primer 16S rRNA -H and 16S rRNA -L of yet another myctophid *Protomyctophum crockeri* (PCMB) designed were such as (SEQ ID NOS 37-38):

16S-H	5'	CGT GTT CTG ATG ATG ATG TGC T	3'
16S-L	5'	ATT CCT TCC TCT TAG TAT G	3'

Page 17, first paragraph:

The sequences of the species specific primer 12S rRNA -H and 12S rRNA -L of yet another myctophid *Protomyctophum crockeri* (PCMB) designed were such as (SEQ ID NOS 39-40):

12S-H	5'	GCT GAA CTT ACT ATG CCC TAC T	3'
12S-L	5'	CCG ATT GAC GCC GAA CTA TG	3'

Page 17, paragraph entitled **Table1**:

**Table1** - Forward primer (SEQ ID NO: 18) designed for cytochrome b gene of *Stenobrachius leucopsarus* ( slmb primer cyt L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 17, paragraph entitled **Table 2**:

**Table 2 -** Backward primer (SEQ ID NO: 17) designed for cytochrome b gene of *Stenobrachius leucopsarus* (slmb primer cyt H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 17, paragraph entitled **Table 3:**

**Table3 -** Forward primer (SEQ ID NO: 20) designed for Internal Transcribed Spacer (ITS2) of *Stenobrachius leucopsarus* (slmb primer ITS2 F) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 17, paragraph entitled **Table 4:**

**Table 4 -** Backward primer (SEQ ID NO: 19) designed for Internal Transcribed Spacer (ITS2) *Stenobrachius leucopsarus* (slmb primer ITS2-H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 17, paragraph entitled **Title 5:**

**Table 5 -** Forward primer (SEQ ID NO: 21) designed for mitochondrial Control region d-Loop of *Stenobrachius leucopsarus* (slmb primer pro-L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 17, paragraph entitled **Table 6:**

**Table 6 -** Backward primer (SEQ ID NO: 22) designed for mitochondrial Control region d-Loop *Stenobrachius leucopsarus* (slmb primer D loop -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 17, paragraph entitled **Table 7:**

**Table 7 -** Forward primer (SEQ ID NO: 23) designed for Rhodopsin gene region of *Stenobrachius leucopsarus* (slmb primer ROD-L) with 5' to 3' end

sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 18, paragraph entitled **Table 8**:

**Table 8** - Backward primer (SEQ ID NO: 24) designed for Rhodopsin gene region of *Stenobrachius leucopsarus* (slmb primer ROD -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 18, paragraph entitled **Table 9**:

**Table 9** - Forward primer (SEQ ID NO: 26) designed for mitochondrial 16S ribosomal RNA region of *Lampanyctus regalis* (LRMB primer 16 S-L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 18, paragraph entitled **Table 10**:

**Table 10** - Backward primer (SEQ ID NO: 25) designed for mitochondrial 16S ribosomal RNA region of *Lampanyctus regalis* (LRMB primer 16 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 18, paragraph entitled **Table 11**:

**Table 11** - Forward primer (SEQ ID NO: 28) designed for mitochondrial 12 S ribosomal RNA region of *Lampanyctus regalis* (LRMB primer 12 S-L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 18, paragraph entitled **Table 12**:

**Table 12** - Backward primer (SEQ ID NO: 27) designed for mitochondrial 12 S ribosomal RNA region of *Lampanyctus regalis* (LRMB primer 12 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.



Page 18, paragraph entitled **Table 13:**

**Table 13 -** Backward primer (SEQ ID NO: 29) designed for mitochondrial 16 S ribosomal RNA region of *Diaphus theta* (DTMB primer 16 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 18, paragraph entitled **Table 14:**

**Table 14 -** Forward primer (SEQ ID NO: 30) designed for mitochondrial 16 S ribosomal RNA region of *Diaphus theta* (DTMB primer 16 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 18, paragraph entitled **Table 15:**

**Table 15 -** Backward primer (SEQ ID NO: 31) designed for mitochondrial 12 S ribosomal RNA region of *Diaphus theta* (DTMB primer 12 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 18, paragraph entitled **Table 16:**

**Table 16 -** Forward primer (SEQ ID NO: 32) designed for mitochondrial 12 S ribosomal RNA region of *Diaphus theta* (DTMB primer 12 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 19, paragraph entitled **Table 17:**

**Table 17 -** Backward primer (SEQ ID NO: 33) designed for mitochondrial 16 S ribosomal RNA region of *Tarletonbeania crenularis* (TCMB primer 16 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 19, paragraph entitled **Table 18:**

**Table 18 -** Forward primer (SEQ ID NO: 24) designed for mitochondrial 16 S ribosomal RNA region of *Tarletonbeania crenularis* (TCMB primer 16 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 19, paragraph entitled **Table 19:**

**Table 19 -** Backward primer (SEQ ID NO: 35) designed for mitochondrial 12 S ribosomal RNA region of *Tarletonbeania crenularis* (TCMB primer 12 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 19, paragraph entitled **Table 20:**

**Table 20 -** Forward primer (SEQ ID NO: 36) designed for mitochondrial 12 S ribosomal RNA region of *Tarletonbeania crenularis* (TCMB primer 12 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 19, paragraph entitled **Table 21:**

**Table 21 -** Backward primer (SEQ ID NO: 37) designed for mitochondrial 16 S ribosomal RNA region of *Protomyctophum crockeri* (PCMB primer 16 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 19, paragraph entitled **Table 22:**

**Table 22 -** Forward primer (SEQ ID NO: 38) designed for mitochondrial 16 S ribosomal RNA region of *Protomyctophum crockeri* (PCMB primer 16 S -L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 19, paragraph entitled **Table 23:**

**Table 23 -** Backward primer (SEQ ID NO: 39) designed for mitochondrial 12 S ribosomal RNA region of *Protomyctophum crockeri* (PCMB primer 12 S

-H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Pages 19-20, paragraph entitled **Table 24:**

**Table 24** - Forward primer (SEQ ID NO: 40) designed for mitochondrial 12 S ribosomal RNA region of *Protomyctophum crockeri* (PCMB primer 12 S - L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 20, paragraph entitled **Table 25:**

**Table 25** - Backward primer (SEQ ID NO: 15) designed for mitochondrial 16 S ribosomal RNA region of *Stenobrachius leucopsarus* (SLMB primer 16 S - H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 20, paragraph entitled **Table 26:**

**Table 26** - Forward primer (SEQ ID NO: 16) designed for mitochondrial 16 S ribosomal RNA region of *Stenobrachius leucopsarus* (SLMB primer 16 S - L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 20, paragraph entitled **Table 27:**

**Table 27** - Backward primer (SEQ ID NO: 13) designed for mitochondrial 12 S ribosomal RNA region of *Stenobrachius leucopsarus* (SLMB primer 12 S -H) with 5' to 3' end sequences (ANTISENSE) and summaries of oligonucleotide and structural analyses.

Page 20, paragraph entitled **Table 28:**

**Table 28** - Forward primer (SEQ ID NO: 14) designed for mitochondrial 12 S ribosomal RNA region of *Stenobrachius leucopsarus* (SLMB primer 12 S - L) with 5' to 3' end sequences (SENSE) and summaries of oligonucleotide and structural analyses.

Page 24, paragraph entitled **Example 6:**

**Example 6:** PCR amplification using forward and backward D-Loop primers of *Stenobrachius leucopsarus*.

The PCR master mix (100 µl) comprised of Taq Buffer MgCl<sub>2</sub> free (10.0 µl), dNTP all the four nucleotides in the ratio of 1:1:1:1 (08.0 µl); D-Loop forward primer 01.0 µl with sequences ( PRO-L : 5' CTA CC 3' ), D-Loop backward 01.0 µl, with sequences (D-Loop H: 5' CCT GAA GTA GGA ACC AGA TG 3') (SEQ ID NO: 4); MgCl<sub>2</sub> ( 01.0 µl); Taq Polymerase (0.5 µl ); and ultrapure water ( 78.2 µl).

Pages 24-25, paragraph entitled **Example 7:**

**Example 7:**

As given in example 6, PCR amplification master mix was prepared using forward and backward 12 S RNA primers; 16 S RNA primers, Cyt b primers; ROD, ITS2 primers and DNA 0.3 µl of *Stenobrachius leucopsarus* was added individually to all tubes and amplified. The primers used were ROD-F: (SEQ ID NO: 8) 5' CAT ATG AAT ACC CTC AGT ACT ACC 3' and ROD-R: (SEQ ID NO: 7) 5' TCT TTC CGC AGC ACA ACG TGG 3' for Rhodopsin DNA probe; 16SBR-H (SEQ ID NO: 12) 5' CCG GTC TGA ACT CAG ATC ACG T 3' and 16SAR-L (SEQ ID NO: 11) 5' CGC CTG TTT ATC AAA AAC AT 3' 16S for 16 S RNA gene probe; 12SA-L: (SEQ ID NO: 9) 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3' and 12SB-L: (SEQ ID NO: 10) 5' AGA GTG ACG GGC GGT GTG T 3' for 12S RNA gene probe and run for 35 cycles in DNA thermo cycler. (Each cycle consisted of 94 degree C for 45 Seconds, 48 degree for 45 seconds, and 72 degree C for 1 minute) and hold at 4 degree Centigrade.

Page 25, paragraph entitled **Example 8:**

**Example 8:**

Cytochrome b DNA probe was amplified by using Cyt 1: (SEQ ID NO: 1) 5' TGA YTT GAA RAA CCA YCG TTG 3' and Cyt 2: (SEQ ID NO: 2) 5' CTC CAR TCT TCG RYT TAC AAG 3' primers followed by reamplification by using CBI-L

(SEQ ID NO: 3) 5' CCA TCC AAC ATC TCA GCA TGA TGA AA 3' and Cyt 2: (SEQ ID NO: 2) 5' CTC CAR TCT TCG RYT TAC AAG 3' primers. The DNA template was of *Stenobranchius leucopsarus* and run for 35 cycles in DNA thermo cycler. (Each cycle consisted of 94°C for 45 Seconds, 48 degree for 45 seconds, and 72 degree C for 1 minute) and hold at 4 degree Centigrade.

Page 25, paragraph entitled **Example 9:**

**Example 9:**

Similarly , primer ITS1F-ITS2R of Internal transcribed spacers was used for the nested PCR's ( ITS1-F: (SEQ ID NO: 41) 5' TTG TAC ACA CCG CCC GTC GC 3' and ITS2-R: (SEQ ID NO: 6) 5' ATA TGC TTA AAT TCA GCG GG 3') and amplified by PCR. Later the ITS2 was reamplified using primers ITS2-F: (SEQ ID NO: 5) 5' CTA CGC CTG TCT GAG TGT C 3' and ITS2-R: (SEQ ID NO: 6) 5' ATA TGC TTA AAT TCA GCG GG 3'. The DNA template was of *Stenobranchius leucopsarus* myctophid fish and run for 35 cycles in DNA thermo cycler. (Each cycle consisted of 94°C for 45 Seconds, 48°C for 45 seconds, and 72°C for 1 minute) and hold at 4 degree Centigrade.

Page 29, paragraph entitled **Example 20:**

**Example 20 :**

PCR for confirmation that transformed bacteria has the plasmids with the D-Loop gene inserts.:

PCR amplification using forward and backward D-Loop primers of *Stenobranchius leucopsarus*.

The PCR master mix (100 µl ) comprised of Taq Buffer MgCl<sub>2</sub> free (10.0 µl ), dNTP all the four nucleotides in the ratio of 1:1:1:1 (08.0 µl); D-Loop forward primer 01.0 µl with sequences ( PRO-L : 5' CTA CC 3'), D-Loop backward 01.0 µl, with sequences (D-Loop H: 5' CCT GAA GTA GGA ACC AGA TG 3') (SEQ ID NO: 4); MgCl<sub>2</sub> (01.0 µl); Taq Polymerase (0.5 µl ); and ultrapure water ( 78.2 µl).

**Marked up rewritten claims:**

9. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for amplification and detection of Cyt b gene contains oligonucleotides with the sequences (SEQ ID NOS 1-2):

CYT 1: 5' TGA YTT GAA RAA CCA YCG TTG 3'  
CYT 2: 5' CTC CAR TCT TCG RYT TAC AAG 3'

10. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for reamplification and detection of Cyt b gene contains oligonucleotides with the sequences (SEQ ID NOS 3 and 2):

CBI-L: 5' CCA TCC AAC ATC TCA GCA TGA TGA AA 3'  
CYT 2: 5' CTC CAR TCT TCG RYT TAC AAG 3'

11. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification and detection of D-Loop gene contains oligonucleotides with the sequences :

PRO-L : 5' CTA CC 3'  
D-LOOP H: 5' CCT GAA GTA GGA ACC AGA TG 3' (SEQ ID NO: 4)

12. (Amended) A method as claimed in claim 1 wherein the forward and backward primers used for PCR amplification of ITS2 gene were

ITS1 F : 5' TTG TAC ACA CCGCCCGTC GC 3' (SEQ ID NO: 41)  
ITS2 R : 5' ATA TGC TTA AAT TCA GCG GG 3' (SEQ ID NO: 6)

13. (Amended) A method as claimed in claim 1 wherein the forward and backward primers used for PCR reamplification of ITS2 gene from ITS1 F and ITS2 R PCR amplification were

ITS2 F: 5' CTA CGC CTG TCT GAG TGT C 3' (SEQ ID NO: 5)  
ITS2 R: 5' ATA TGC TTA AAT TCA GCG GG 3' (SEQ ID NO: 6)

14. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of Rhodopsin gene Rod contains oligonucleotides with the sequences (SEQ ID NOS 8 and 7):

ROD-F: 5' CAT ATG AAT ACC CTC AGT ACT ACC 3'  
 ROD-R: 5' TCT TTC CGC AGC ACA ACG TGG 3'

15. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of 12S RNA gene contains oligonucleotides with the sequences (SEQ ID NOS 9-10):

12 SA-L: 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3'  
 12 SB-H : 5 ' AGA GTG ACG GGC GGT GTG T 3'

16. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of 16S RNA gene contains oligonucleotides with the sequences:

16 SAR -L: 5' CGC CTG TTT ATC AAA AAC AT 3' (SEQ ID NO: 11)  
 16 SBR-H : 5 ' CCG GTC TGA ACT CAG ATC ACG T 3' (SEQ ID NO: 12)

17. (Amended) A method as claimed in claim 1 wherein the forward and backward primers used for PCR amplification of Rhodopsin gene Rod were (SEQ ID NOS 8 and 7):

ROD-F: 5' CAT ATG AAT ACC CTC AGT ACT ACC 3'  
 ROD-R: 5' TCT TTC CGC AGC ACA ACG TGG 3'

18. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of 12S RNA gene were (SEQ ID NOS 9-10):

12 SA-L: 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3'  
 12 SB-H: 5 ' AGA GTG ACG GGC GGT GTG T 3'

19. (Amended) A method as claimed in claim 1 wherein the primer set (forward and backward primers) used for PCR amplification of 16S RNA gene were (SEQ ID NOS 11-12):

16 SAR -L: 5' CGC CTG TTT ATC AAA AAC AT 3'  
 SBR-H: 5 ' CCG GTC TGA ACT CAG ATC ACG T 3'

29. (Amended) A method claimed in claim 1 wherein the cycle sequencing primer for CYT b gene consisted of oligonucleotides with the sequence:

CYT 1: 5' TGA YTT GAA RAA CCA YCG TTG 3' (SEQ ID NO: 1)

30. (Amended) A method claimed in claim 1 wherein the cycle sequencing primer for CYT b gene consisted of oligonucleotides with the sequence:

CYT 2: 5' CTC CAR TCT TCG RYT TAC AAG 3' (SEQ ID NO: 2)

31. (Amended) A method claimed in claim 1 wherein the cycle sequencing primer for CYT b gene consisted of oligonucleotides with the sequence:

CBI-L: 5' CCA TCC AAC ATC TCA GCA TGA TGA AA 3' (SEQ ID NO: 3)

33. (Amended) A method claimed in claim 1 wherein the backward cycle sequencing primer for D-Loop region consisted of oligonucleotides with the sequence:

D-LOOP H: 5' CCT GAA GTA GGA ACC AGA TG 3' (SEQ ID NO: 4)

34. (Amended) A method as claimed in claim 1 wherein the forward primer used for cycle sequencing of ITS2 gene consisted of oligonucleotides with the sequence:

ITS 1 -F : 5' TTG TAC ACA CCG CCC GTC GC 3' (SEQ ID NO: 41)

35. (Amended) A method as claimed in claim 1 wherein the backward primer used for cycle sequencing of ITS2 gene consisted of oligonucleotides with the sequence:

ITS2 -R : 5' ATA TGC TTA AAT TCA GCG GG 3' (SEQ ID NO: 6)

36. (Amended) A method as claimed in claim 1 wherein the forward primer used for cycle sequencing of Rhodopsin gene Rod consisted of oligonucleotides with the sequence:



ROD-F: 5' CAT ATG AAT ACC CTC AGT ACT ACC 3' (SEQ ID NO: 8)

37. (Amended) A method as claimed in claim 1 wherein the backward primer used for cycle sequencing consisted of oligonucleotides with the sequence:

ROD-R: 5' TCT TTC CGC AGC ACA ACG TGG 3' (SEQ ID NO: 7)

38. (Amended) A method as claimed in claim 1 wherein the forward primer used for cycle sequencing of 12S RNA gene consisted of oligonucleotides with the sequence:

12 SA-L: 5' AAA CTG GGA TTA GAT ACC CCA CTA T 3' (SEQ ID NO: 9)

39. (Amended) A method as claimed in claim 1 wherein the backward primer used for cycle sequencing of 12S RNA gene consisted of oligonucleotides with the sequence:

12 SB-H: 5' AGA GTG ACG GGC GGT GTG T 3' (SEQ ID NO: 10)

40. (Amended) A method as claimed in claim 1 wherein the forward primer used for cycle sequencing of 16S RNA gene consisted of oligonucleotides with the sequence:

16 SAR -L: 5' CGC CTG TTT ATC AAA AAC AT 3' (SEQ ID NO: 11)

41. (Amended) A method as claimed in claim 1 wherein the backward primer used for cycle sequencing of 16S RNA gene consisted of oligonucleotides with the sequence:

16 SBR-H: 5' CCG GTC TGA ACT CAG ATC ACG T 3' (SEQ ID NO: 12)

74. (Amended) The nucleotide base sequences of PSL CYTL [( 748 bp)] (747 bp) comprising (SEQ ID NO: 42):

5'

CTTNCCCATT	TTGGGCGCTT	NGGCNCGCTN	CTCCNCGAGA	CTCTGCGTAN
TAATCCAANT	CNCTNCGGGC	CNCTCCCTAC	CANTNCNCTA	CACCNCAAAT
TNCAACCCNG	TTTCCTCATC	ANTCAACCAC	ATCTGTCGAA	AACNTCAACT
ACGGCTGACT	AATCCGAAAA	CATGCACGCT	AACGGTGCCT	CTTTCTTCTT
CATCTGTATT	TATCTNCNCN	TTGGANGAGG	ACTATNCTAC	GGATCCTACC
TCTACGAAGA	GACGTGAGGT	GTTGGTGTTA	TTCTTCTCCT	TCTAATAATG
ATGACTGCNT	TTGTTGGCTA	TGTGCTNCCC	NGAGGACAAA	TGTCCTTTTG
AGGTGCTACT	GTCATTACAA	NCCTACTCTC	TGCTGTNCCG	TNTGTTNGCG
GCNCTCTANT	TCAATGAATT	TGAGGTGGCT	TCTCCGTAAA	CACGCAACGC
TCACTCGTTT	CTTCGCNTTC	CACTTCTTGT	TCCCATTGTG	TGTCGCNGCT
ATAACCNNGG	TTCACCNAT	TTNCCGACAT	CAAACAGGCT	CTAAANCCCC
CCCGGNTTGA	CTCCATACAA	CAAAACCCTC	CACCCTATTC	NCTATAAAAC
TCTAGGTTTCG	TGCCCCGTATT	GGCTTACTTC	ATGNCTATTT	CCCNGNCGGA
GGGACNAAAA	TTCCTGCACC	CCCTCCCCNC	AAAATAAANA	ATGTGTCTNT
CCTACCANAA	AACAACNNAN	ACGGGGTNTG	CNCTTCCATC	ATCCACN 3'

75. (Amended) The nucleotide base sequences of PSLITS2F comprises:  
(225BP) (SEQ ID NO: 43)

5'				
TCTACGATCT	ACCGGCNTTT	NNTGTGGAAA	GACGATCATG	
CATTTATGTG	TGTCTTTCTA	TGGATTTGAA	CCGTGTGGTA	
CGTCTTTGCG	TACTGCTTGG	AAGGCTCAAC	TTGCTTCTGT	
CCTTCTCTTG	CAGTCTCGCA	CTGTCTATGC	AACGTGTTCT	
ACTTCGACTT	CTGTCGAAAA	ATCTTACTTT	TGACCTCAGA	
TCAGACAAGA	CTACCCGCTG	AATTT	3'	

76. (Amended) The nucleotide base sequences of PSL PROL  
comprises: [(749 BP)] (750 BP) (SEQ ID NO: 44)

5'				
CCTTTTCGGN	ATAGGCCCAN	CTCAAATGAA	TTCCTTCTCT	
CCTGGTCCAA	GCCCCAACTG	TGGACGGCAG	GTTGACAATG	
GTTACAAATC	GTGACAAATC	GGCTACATAA	TTGCCGATAG	
CGATGTCGTC	AAACCAAGTC	AAACAATGGC	CGATGTATAT	

CGGCCAAACC	CATATATGGG	TCTGGCTGTA	GTTTGTGTTG
AGCAACGTCA	CACCAGTGTC	TGGTCAGCAT	ATAAGATGTT
GACATCTTGC	AACATCTTAC	CCACAGACAG	ACAGTTACGG
CTGCTTACGA	ANGGCGCTAG	TGTTGTGGTG	AGAAACGAAG
ATACATACGT	CAAACAGACG	CCGGTGCACT	TGAAGACACT
GTTTGAAGGT	GCCGCACTAC	TTGACAGACA	GCCCATGATG
CGCTGGACAG	TGACCAAAGC	TACNGGAGGA	CCANATGGAA
ATCCTGTTGG	CGTTGCCGTG	GGACTCAAGT	TGTACACTTT
TGGATGGTTG	ATCACTANAN	CCGCTGCCGG	GAGAAGCACT
CGCTCCTGGT	TCACTAATCA	GATTGAGGTT	AACCANATTG
ANGTAAACAT	CTTCAACACA	GTGTCTTTAT	GCTGGATGAA
ATTNAGCCCA	CNGGACACCA	NAAAAGAATT	NCCNCTGGTT
CTNNCGGGGG	NCCCCNNNAA	CGNNTNTTCC	CCTTNTCTCN
NNNGCGGNGA	AGTTNCCCCC	CCCCACTNAN	NTCTTCCTTC
AANANNTTTC	CNCCNNNAGA	GGTTTTCCCN	3'

77. (Amended) The nucleotide base sequences of ROD PSL SLMB comprises: (748 BP) (SEQ ID NO: 45)

5'			
CCTGGTAGGG	TTCCCCGTCA	ACTTCCTCAC	ACTGTACCTC
ACNTTCGAGC	ACAAGAAGCT	ACTAACCCCC	TTAAACTACA
TCCTGCTCAA	CCTGGCGGTC	GGAGACCTCC	TGATGGTGTA
AGGAGGGTTC	ACCACCACCA	TCTACACCTC	CATGCACGGC
TACTTCGTCC	TAGGGAAACT	GGGCTGCGCC	ATCGAAGGTT
TCATGGCCAC	CCATGGTGGT	CAGGTCGCCC	TTTGGTCCCT
GGTGGTTTTG	GCCGTGGAAA	GGTGGCTGGT	CGTCTGCAAN
CCCATCTCCA	GCTTCCGCTT	CCAGGAGTCC	CACTCCCTCA
TGGGCCTGGC	CGTGACCTGG	GTGATGGCGA	CGGCTTGTTT
TGTGCCCCCC	CTGGGTCGGC	TGGTCTCGCT	ACATCCCAGA
AGGCATGCAG	TGCTCATGCG	GAATGGACTA	CTACACTCCC
GCGCCGGGCG	TCAACAATGA	ATCCTACGTN	GTGTACATGT
TCNTCANAAA	AANAATNGGA	CCNCNGGGCG	ATCATNTTGN

TANGNNAAGG	CCAGNTGNTG	NGAGCAGTCA	AGGCGGCCGC
CGCCGCCCAG	CAAGAGTCCG	AGACCACCCA	GAGGGCCGAG
AGGGAAGTCA	CCCGNATGGT	NATNANGATG	GTNATNTCNT
TCNTGGTAAG	NAGGGNGCCA	NACGCCAGCG	TGGCCTGGTG
GATCTTNGN	AACCAGGGNG	CAGAATTAGG	CCCNGTNTTC
ATGACCCTGC	CGGCNTTCTT	TGCCAAGA	3'

78. (Amended) A method as claimed in claim 1 wherein FORWARD ( L) primers of CYT b gene region for myctophid fish *Stenobrachius leucopsarus* is an oligonucleotide comprising (SEQ ID NO: 18):

5' CAA CCT CAT CTG TCG TAA AC 3'

and having the following characteristics:

- i. is a 20-mer DNA oligonucleotide ( sense),
- ii. has melting temperature of 56.4 degree celcius,
- iii. has a molecular weight of 6101.0,
- iv. has no hairpin loops,
- v. has no single dimers,
- vi. has no other dimers,
- vii. has no single bulge loops or internal loops, and
- viii. has no palindromes.

79. (Amended) A method as claimed in claim 1 wherein BACKWARD (H) primer of CYT b gene region for myctophid fish *Stenobrachius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 17):

5' GCT CGG GCT GCT GGA ATC TT 3'

and having the following characteristics:

- i. is a 20-mer DNA
- ii. is an antisense oligonucleotide
- iii. has a melting point of 70.8 degree celcius.
- iv. has a molecular weight of 6220.1.
- v. has no hairpin loops, no single bulge loops, no other internal loops, no single internal loops, no other bulge loops or palindromes.
- vi. no single dimers or other dimers.

80. (Amended) A method as claimed in claim 1 wherein forward primer of ITS2 F gene region for myctophid fish *Stenobranchius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 20):

5' ACT TGA CTG ACC TTC TTA CT 3'

and having the following characteristics:

- i. is a 20-mer sense oligonucleotide,
- ii. has a melting point of 51.3 degree celcius,
- iii. has a molecular weight of 6098.0,
- iv. has no palindromes, loops and dimers,

81. (Amended) A method as claimed in claim 1 wherein forward primer of ITS2 H gene region for myctophid fish *Stenobranchius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 19):

5' ATA CTC TGC GGA CAT ACT TGA CTG 3'

and having the following characteristics:

- i. is a 24-mer antisense oligonucleotide,
- ii. has a melting point of 65.4 degree celcius.
- iii. has a molecular weight of 7407.9.
- iv. has no palindromes, loops and dimers.

82. (Amended) A method as claimed in claim 1 wherein forward primer of pro-L for myctophid fish *Stenobranchius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 21):

5' CAG TCT CGT CAA ACC AAG TCA AAC 3'

and having the following characteristics:

- i. is a 24-mer sense oligonucleotide
- ii. has a melting point of 67.8 degree celcius.
- iii. has a molecular weight of 7354.9.
- iv. has no palindromes, loops and dimers.

83. (Amended) A method as claimed in claim 1 wherein backward primer for Dloop for mitochondrial control region (dloop H) gene region for myctophid fish *Stenobranchius leucopsarus* is an oligonucleotide comprising (SEQ ID NO: 22):

5' ATA ATC ATC CAG CAT AAA CAC AC 3'

and having the following characteristics:

- i. is a 23-mer antisense oligonucleotide,
- ii. has a melting point of 61.2 degree celcius.
- iii. has a molecular weight of 7033.7.
- iv. has no palindromes, loops and dimers.

84. (Amended) A method as claimed in claim 1 wherein the FORWARD primer ( ROD- L) for Rhodopsin gene region of myctophid fish *Stenobranchius leucopsarus* is an oligonucleotide comprising (SEQ ID NO: 23):

5' CCT GGT AGA GTT CGC CGT CA 3'

and having the following characteristics:

- i. is a 20-mer sense oligonucleotide
- ii. has a melting point of 67.4 degree celcius.
- iii. has a molecular weight of 6189.0.
- iv. has no palindromes, loops and dimers.

85. (Amended) A method as claimed in claim 1 wherein the backward primer ( ROD- H) for Rhodopsin gene region of myctophid fish *Stenobranchius leucopsarus* is an oligonucleotide comprising (SEQ ID NO: 24):

5' CGT GTT CCT TAT CAT TGT GCC T 3'

and having the following characteristics:

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 66.4 degree celcius.
- iii. has a molecular weight of 6738.4.
- iv. has no palindromes, loops and dimers.

86. (Amended) A method as claimed in claim 1 wherein the forward primer of 16S-L of the myctophid fish *Lampanyctus regalis* is an oligonucleotide comprising (SEQ ID NO: 26):

5' CAC CAG CCA AGT ATG TTT CTC 3'

and having the following characteristics:

- i. is a 21-mer sense oligonucleotide
- ii. has a melting point of 61.5 degree celcius.
- iii. has a molecular weight of 6421.4.
- iv. has no palindromes, loops and dimers.

87. (Amended) A method as claimed in claim 1 wherein the backward primer of 16s rRNA of myctophid fish *Lampanyctus regalis* is an oligonucleotide comprising (SEQ ID NO: 25):

5' TCG TAG TTC AGC AGT CAG 3'

and having the following characteristics:

- i. is a 18-mer antisense oligonucleotide
- ii. has a melting point of 51.2 degree celcius.
- iii. has a molecular weight of 5594.7.
- iv. has no palindromes, hairpin loops and dimers.

88. (Amended) A method as claimed in claim 1 wherein the forward primer 16S-L of myctophid fish *Lampanyctus regalis* is an oligonucleotide comprising (SEQ ID NO: 28):

5' CTA TTC GCC TCG CTC AGA C 3'

and having the following characteristics:

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 62.1 degree celcius.
- iii. has a molecular weight of 5779.8.
- iv. has no palindromes, hairpin loops and dimers.

89. (Amended) A method as claimed in claim 1 wherein a primer 12S-H for *Lampanyctus regalis* (LRMB) is an oligonucleotide comprising (SEQ ID NO: 27):

5' GCC TCC ATC ATC CCT CAC CTT AC 3'

and having the following characteristics:

- i. is a 23-mer antisense oligonucleotide
- ii. has a melting point of 70.8 degree celcius.

- iii. has a molecular weight of 6895.5
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

90. (Amended) A method as claimed in claim 1 wherein the primer 12S-L for *Lampanyctus regalis* (LRMB) is an oligonucleotide comprising (SEQ ID NO: 28):

5' CTA TTC GCC TCG CTC AGA C 3'

and having the following characteristics:

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 62.1 degree celcius.
- iii. has a molecular weight of 5779.8
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

91. (Amended) A method as claimed in claim 1 wherein 16S-L forward primer for *Diaphus theta* (DTMB) is an oligonucleotide comprising (SEQ ID NO: 30):

5' AAA TCC GCC CTT ATG TGT GTT C 3'

and having the following characteristics:

- i. is a 22-mer sense oligonucleotide
- ii. has a melting point of 67.9 degree celcius.
- iii. has a molecular weight of 6756.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

92. (Amended) A method as claimed in claim 1 wherein 16S-H backward primer for *Diaphus theta* (DTMB) is an oligonucleotide comprising (SEQ ID NO: 29):

5' CTC CGT CCG TCT CGC CTC TG 3'

and having the following characteristics:

- i. is a 20-mer antisense oligonucleotide
- ii. has a melting point of 71.7 degree celcius.
- iii. has a molecular weight of 6052.0
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.



93. (Amended) A method as claimed in claim 1 wherein 12S-H forward primer for *Diaphus theta* (DTMB) is an oligonucleotide comprising (SEQ ID NO: 31):

5' CAT CGG CTT GCT CTA TTC CTT G 3'

and having the following characteristics:

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 68.8 degree celcius.
- iii. has a molecular weight of 6723.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

94. (Amended) A method as claimed in claim 1 wherein 12S-L forward primer for *Diaphus theta* (DTMB) is an oligonucleotide comprising (SEQ ID NO: 32):

5' TCT ATC GGC GGC GTA TCA C 3'

and having the following characteristics:

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 65.8 degree celcius.
- iii. has a molecular weight of 5859.8
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

95. (Amended) A method as claimed in claim 1 wherein 16S-H primer for *Tarletonbeania crenularis* (TCMB) is an oligonucleotide comprising (SEQ ID NO: 33):

5' GGC GAT TCT ACG GCA CGG GCG 3'

and having the following characteristics:

- i. is a 21-mer antisense oligonucleotide
- ii. has a melting point of 80.4 degree celcius.
- iii. has a molecular weight of 6568.3
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

96. (Amended) A method as claimed in claim 1 wherein 16S-L forward primer for *Tarletonbeania crenularis* (TCMB) is an oligonucleotide comprising (SEQ ID NO: 34):

5' AAA CTG GTC CTC AAC TAT GTC A 3'

and having the following characteristics:

- i. is a 22-mer sense oligonucleotide
- ii. has a melting point of 60.7 degree celcius.
- iii. has a molecular weight of 6758.5
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

97. (Amended) A method as claimed in claim 1 wherein 16S-H backward primer for *Tarletonbeania crenularis* (TCMB) is an oligonucleotide comprising (SEQ ID NO: 33):

5' GGC GAT TCT ACG GCA CGG GCG 3'

and having the following characteristics:

- i. is a 21-mer antisense oligonucleotide
- ii. has a melting point of 80.4 degree celcius.
- iii. has a molecular weight of 6568.3
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

98. (Amended) A method as claimed in claim 1 wherein 12S-H backward primer for *Tarletonbeania crenularis* (TCMB) is an oligonucleotide comprising (SEQ ID NO: 35):

5' CCG ATT CAG CCA CGA TTC CCT C 3'

and having the following characteristics:

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 74.6 degree celcius.
- iii. has a molecular weight of 6671.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

99. (Amended) A method as claimed in claim 1 wherein 12S-L forward primer for *Tarletonbeania crenularis* (TCMB) is an oligonucleotide comprising (SEQ ID NO: 42):

5' CCT AAA GCC CAG ATA ACT ACA 3'

- i. is a 21-mer sense oligonucleotide
- ii. has a melting point of 59.2 degree celcius.
- iii. has a molecular weight of 6432.3
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

100. (Amended) A method as claimed in claim 1 wherein 16S-H backward primer for *Protomyctophum crockeri* (PCMB) is an oligonucleotide comprising (SEQ ID NO: 37):

5' CGT GTT CTG ATG ATG ATG TGC T 3'

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 64.7 degree celcius.
- iii. has a molecular weight of 6867.5
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

101. (Amended) A method as claimed in claim 1 wherein 16S-L forward primer for *Protomyctophum crockeri* (PCMB) is an oligonucleotide comprising (SEQ ID NO: 38):

5' ATT CCT TCC TCT TAG TAT G 3'

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 49.5 degree celcius.
- iii. has a molecular weight of 5799.8
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

102. (Amended) A method as claimed in claim 1 wherein 12S-H backward primer for *Protomyctophum crockeri* (PCMB) is an oligonucleotide comprising (SEQ ID NO: 39):

5' GCT GAA CTT ACT ATG CCC TAC T 3'

- i. is a 22-mer antisense oligonucleotide
- ii. has a melting point of 60.3 degree celcius.
- iii. has a molecular weight of 6725.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

103. (Amended) A method as claimed in claim 1 wherein 12S-L forward primer for *Protomyctophum crockeri* (PCMB) is an oligonucleotide comprising (SEQ ID NO: 40):

5' CCG ATT GAC GCC GAA CTA TG 3'

- i. is a 20-mer sense oligonucleotide
- ii. has a melting point of 68.1 degree celcius.
- iii. has a molecular weight of 6182.1
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

104. (Amended) A method as claimed in claim 1 wherein 16S-H backward primer for *Stenobrachius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 15):

5' TAC GCA TAA CGG CTC TGG 3'

- i. is a 18-mer DNA oligonucleotide ( Antisense)
- ii. has a melting point of 61.4 degree celcius.
- iii. has a molecular weight of 5579.7
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

105. (Amended) A method as claimed in claim 1 wherein 16S-L forward primer for *Stenobrachius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 16):

5' CTA CTA CAC CTC AAC TAC ATC T 3'

- i. is a 22-mer sense oligonucleotide
- ii. has a melting point of 52.4 degree celcius.

- iii. has a molecular weight of 6638.4
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

106. (Amended) A method as claimed in claim 1 wherein 12S-H forward primer for *Stenobrachius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 13):

5' CCC ACT CAC TGC TAA CTC C 3'

- i. is a 19-mer sense oligonucleotide
- ii. has a melting point of 58.4 degree celcius.
- iii. has a molecular weight of 5708.8
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.

107. (Amended) A method as claimed in claim 1 wherein 12S-L forward primer for *Stenobrachius leucopsarus* (SLMB) is an oligonucleotide comprising (SEQ ID NO: 14):

5' GGC TAA CTA CAA TCA TCT GCT 3'

- i. is a 21-mer sense oligonucleotide
- ii. has a melting point of 58.5 degree celcius.
- iii. has a molecular weight of 6445.2
- iv. has no hairpin loops, dimers, palindromes or bulges and internal loops of either single or other kinds.